

Comparative in-vitro activity of selected new β -lactam antimicrobials against *Neisseria gonorrhoeae**

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SUMMARY Four new β -lactam antimicrobials, ceftriaxone, cefotiam, cefonicid, and mecillinam, were evaluated in vitro against 72 β -lactamase-negative and 26 β -lactamase-positive isolates of *Neisseria gonorrhoeae*. Ceftriaxone was the most active of the antimicrobials tested. It inhibited all isolates, regardless of β -lactamase activity, at a concentration of ≤ 0.015 $\mu\text{g/ml}$. Cefotiam and cefonicid were also active against both groups but not as active as ceftriaxone. Both groups of *N gonorrhoeae* showed a high degree of resistance against mecillinam.

Introduction

The worldwide emergence of β -lactamase-positive strains of *Neisseria gonorrhoeae*¹ and the occasional occurrence of spectinomycin-resistant *N gonorrhoeae* strains² have prompted the search for new antimicrobial agents that might prove effective in treating infections caused by these resistant strains. Among the presently available newer β -lactam antimicrobial agents cefoxitin, cefuroxime, and cefotaxime have excellent in-vitro activity against *N gonorrhoeae*, including β -lactamase-positive strains.³⁻⁵ Recent clinical studies concerning the use of these antimicrobial agents in treating gonococcal infections have shown encouraging results.⁶⁻⁸ This study was conducted to evaluate the in-vitro activity of four new β -lactam antimicrobials, ceftriaxone, cefotiam, cefonicid, and mecillinam, against β -lactamase-negative and β -lactamase-positive strains of *N gonorrhoeae*.

Materials and methods

BACTERIAL STRAINS

A total of 98 strains of *N gonorrhoeae* were tested. Seventy-two β -lactamase-negative strains were

collected during 1980 from patients with anogenital infections at the Hennepin County Medical Center, Minneapolis, Minnesota. Twenty-six β -lactamase-positive strains were obtained from the following sources: W Harrison, Naval Regional Medical Center, San Diego, California; W Hall, Veterans Administration Medical Center, Minneapolis, Minnesota; and the Center for Disease Control, Atlanta, Georgia. The identity of the isolates was confirmed by growth on Thayer-Martin agar, Gram stain, positive oxidase reaction, and acidification of glucose but not of maltose, lactose, or sucrose. The organisms were frozen in tryptic soy broth (Difco Laboratories, Detroit, Michigan) containing 20% glycerol and stored at -80°C . *N gonorrhoeae* isolates were tested for β -lactamase activity by an acidometric method.⁹

ANTIMICROBIALS

Ceftriaxone (RO-139904) and mecillinam (RO-109070) were obtained from Hoffman-La Roche Inc, Nutley, New Jersey; cefotiam (CGP14221/E) from CIBA-Geigy Corporation, Summit, New Jersey; and cefonicid (SK&F 75073) from Smith Kline and French Laboratories, Philadelphia, Pennsylvania. The antimicrobials were supplied as dry powders and stored at -20°C .

SUSCEPTIBILITY TESTING

The minimum inhibitory concentrations (MICs) of ceftriaxone, cefotiam, cefonicid, and mecillinam were determined by the agar dilution technique.¹⁰ Two-fold dilutions of the antimicrobial agents, from

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8 to 0.015 $\mu\text{g/ml}$, were distributed into Mueller-Hinton agar supplemented with 2% haemoglobin and 1% IsoVitalex. The frozen gonococcal isolates were thawed and grown overnight on chocolate agar and then suspended in tryptic soy broth until the turbidity matched that of a 0.5 McFarland standard. One microlitre of a 1/10 dilution of the adjusted suspension (10^4 colony-forming units) was inoculated on to the plates containing the antimicrobial agent with a Steers replicator. The plates were incubated for 18 to 24 hours at 35°C in a CO_2 atmosphere. The minimum inhibitory concentration was defined as the lowest concentration of the antimicrobial agent that inhibited visible growth on the surface of the agar.

Results

The MICs of four antimicrobial agents against the β -lactamase-negative strains of *N. gonorrhoeae* are given in table I. Ceftriaxone inhibited all isolates at a concentration of $\leq 0.015 \mu\text{g/ml}$. Both cefotiam and cefonicid were less active than ceftriaxone. MICs for 90% of the isolates of cefotiam were four-fold and of cefonicid eight-fold higher than those of ceftriaxone. These isolates showed a high degree of resistance to mecillinam.

TABLE I Antimicrobial susceptibility of 72 β -lactamase-negative strains of *N. gonorrhoeae*

Antimicrobial agent	MIC ($\mu\text{g/ml}$)		
	Range	For strains (%)	
		50	90
Ceftriaxone	≤ 0.015	≤ 0.015	≤ 0.015
Cefotiam	≤ 0.015 -1.0	≤ 0.015	0.06
Cefonicid	≤ 0.015 -1.0	0.06	0.125
Mecillinam	0.125-8.0	2.0	4.0

The MICs of the four antimicrobial agents against β -lactamase-positive strains of *N. gonorrhoeae* are shown in table II. Ceftriaxone was also highly effective against this group. All strains were inhibited at a concentration of $\leq 0.015 \mu\text{g/ml}$. Cefotiam and cefonicid were less active than ceftriaxone against these isolates. MICs for 90% of the isolates of cefotiam were eight-fold and of cefonicid 32-fold higher than those of ceftriaxone. This group was also highly resistant to mecillinam.

Discussion

Of the four new β -lactam antimicrobial agents tested in this study, ceftriaxone was found to be the most active against *N. gonorrhoeae* regardless of β -lactamase production. These results agree with the

TABLE II Antimicrobial susceptibility of 26 β -lactamase-positive strains of *N. gonorrhoeae*

Antimicrobial agent	MIC ($\mu\text{g/ml}$)		
	Range	For strains (%)	
		50	90
Ceftriaxone	≤ 0.015	≤ 0.015	≤ 0.015
Cefotiam	≤ 0.015 -0.125	0.03	0.125
Cefonicid	0.06-0.50	0.25	0.50
Mecillinam	2.0- ≥ 8.0	≤ 8.0	≥ 8.0

study of Yoshikawa *et al.*¹¹ in which ceftriaxone was compared with cefuroxime, cefoxitin, and ampicillin against 87 β -lactamase-negative and eight β -lactamase-positive strains of *N. gonorrhoeae*. Ceftriaxone was the most effective of the four antimicrobial agents tested. Cefonicid and cefotiam have also been tested previously against *N. gonorrhoeae*.^{12,13} The results were similar to those obtained in the present study. These two antimicrobial agents are quite active against *N. gonorrhoeae*; however, they are not as active as ceftriaxone. Both cefonicid and cefotiam are presently undergoing clinical evaluation. Their role in the treatment of gonococcal infections remains to be determined. Mecillinam does not appear to be a potentially useful agent for the treatment of gonococcal infections. Although it is a β -lactam antimicrobial like the other three, it shows very poor activity against *N. gonorrhoeae*. To our knowledge, the in-vitro activity of mecillinam against *N. gonorrhoeae* has not previously been reported.

In the present study MICs of ceftriaxone against *N. gonorrhoeae* were similar to those for cefotaxime.^{4,5} Cefotaxime has been shown to be highly effective against gonococcal infections due to β -lactamase-negative and β -lactamase-positive strains of *N. gonorrhoeae*.⁸ Ceftriaxone is presently undergoing clinical trials in the treatment of gonococcal infections. In a preliminary study uncomplicated gonorrhoea in men due to β -lactamase-negative strains was successfully treated with ceftriaxone in single intramuscular doses of 125 mg (15 patients), 250 mg (16 patients), and 500 mg (15 patients).¹⁴ Among the new β -lactam antimicrobials in use or under clinical investigation for gonococcal infections, ceftriaxone appears to be the most promising because of its effectiveness at low doses. Further clinical evaluation of this antimicrobial for single-dose treatment of infections due to both β -lactamase-negative and β -lactamase-positive strains of *N. gonorrhoeae* is warranted.

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